

# **Demi Fraser Broth**

Liquid medium for detection and enumeration of *Listeria monocytogenes* and *Listeria* spp, according to ISO 11290 (both Part 1 and Part 2).

TYPICAL FORMULA	(g/l)
Enzymatic Digest of Animal Tissues	5.0
Enzymatic Digest of Casein	5.0
Meat Extract	5.0
Yeast Extract	5.0
Sodium Chloride	20.0
Disodium Phosphate, Anhydrous	9.6*
Potassium Dihydrogen Phosphate	1.35
Aesculin	1.0
Lithium Chloride	3.0
Nalidixic Acid	0.01
Acriflavine	0.0125
Final pH 7.2 ± 0.2 at 25°C	

<sup>\*</sup>Equivalent to 12.0 g of Disodium Hydrogen Phosphate, Dihydrate.

### **DESCRIPTION**

Demi Fraser Broth is a liquid medium used with supplements for the primary enrichment of *L monocytogenes* and *Listeria* spp from food, animal feeding and environmental samples in the area of food production and food handling. This medium can also be used as a diluent for the preparation of the initial suspension when performing the enumeration procedure.

The medium is completed after the addition of Listeria Fraser Supplement which is available in the following forms:

- Freeze-dried (ref. 81028)
- Liquid (ref. 80304)

Both items contain Ammonium Iron(III) Citrate.

### **PRINCIPLE**

Enzymatic digest of animal tissues, enzymatic digest of casein and meat extract provide nitrogen, vitamins, minerals and amino acids for organisms growth. Yeast extract is a source of vitamins, particularly of B-group. Sodium chloride maintains the osmotic balance of the medium and in a so high concentration inhibits enterococci. Potassium and sodium phosphates act as buffer system. Aesculin is hydrolyzed by all *Listeria* species to aesculetin. Lithium chloride is inhibitory for the accompanying flora. Acriflavine and nalidixic acid are selective agents.

Ferric ions provided by ammonium iron(III) citrate will react with aesculetin producing a blackening of the medium.

### PREPARATION

Suspend 55 g of powder in 1 liter of deionized or distilled water. Bring to boil and shake until completely dissolved. Sterilize at 121°C for 15 minutes. Cool up to 45-50°C. Aseptically, add the content of 2 vials of Listeria Fraser Supplement. NOTE: Either ref. 81028 or ref. 80304 can be chosen. The latter is a ready to use liquid solution whereas ref. 81028 needs to be reconstituted with water before use.

### TECHNIQUE

- Add sample to Demi Fraser Broth to prepare a 10-fold dilution (w/v or v/v).
   Skip directly to step 3 for the enumeration method described in ISO 11290-2.
   For the detection method in ISO 11290-1, incubate at 30 ± 1°C for 25 ± 1 h.
- 2. Transfer 0.1 ml of the primary enrichment culture into 10 ml of Listeria Fraser Broth (ref. 24131). Incubate at  $37 \pm 1^{\circ}$ C for  $24 \pm 2$  h.
- From the enrichment cultures or from the initial suspension (depending on the method used) surface inoculate onto O.A. Listeria Agar (ref. 10620).
  - Incubate at  $37 \pm 1^{\circ}$ C for  $24 \pm 2$  h and for an additional  $24 \pm 2$  h.
- 4. Following the procedure given by ISO 11290-1, use the selective enrichments to inoculate a second selective medium, e.g. Listeria Palcam Agar (ref. 10041), Listeria Oxford Agar (ref. 61016). Refer to the relevant technical sheet for further details.

### INTERPRETATION OF RESULTS

A blackening of Demi Fraser Broth can be observed after incubation.

Blue-green colonies with or without halo on O.A. Listeria Agar are considered presumptive *Listeria* spp. Typical colonies of *L. monocytogenes* are surrounded by an opaque halo. For the enumeration method count all colonies presumed to be *L. monocytogenes* and/or *Listeria* spp.

For confirmation, subculture onto appropriate non-selective agar, e.g. Blood Agar, Nutrient Agar, TSYEA (ref. 10432). Then, carry out confirmation tests including a positive and negative control.

### STORAGE CONDITIONS

The powder is very hygroscopic, store the powder at 10-30°C, in a dry environment, in its original container tightly closed and use it before the expiry date on the label or until signs of deterioration or contamination are evident. Store prepared medium at 2-8°C away from light.





#### WARNING AND PRECAUTIONS

The product does not contain hazardous substances in concentrations exceeding the limits set by current legislation and therefore is not classified as dangerous. It is nevertheless recommended to consult the safety data sheet for its correct use. The product is designed for professional use only and must be used by properly trained operators.

### **DISPOSAL OF WASTE**

Disposal of waste must be carried out according to the national and local regulations in force.

### **REFERENCES**

- ISO 11290-1:2017. Microbiology of the food chain Horizontal method for the detection and enumeration of Listeria monocytogenes and Listeria spp. – Part 1: Detection Method.
- 2. ISO 11290-2:2017. Microbiology of the food chain Horizontal method for the detection and enumeration of *Listeria monocytogenes* and *Listeria* spp. Part 2: Enumeration Method.
- 3. EN ISO 11133:2014. Microbiology of food, animal feed and water Preparation, production, storage and performance testing of culture media.
- 4. Rapporto ISTISAN 96/35. ISSN 1123-3117. Metodi di analisi per il controllo microbiologico degli alimenti.
- 5. Normalisation Française, AFNOR (1993) V08-55.
- 6. Fraser. J.A and Sperber W.H (1988) J. Food Prot , 51, 762-765.



## **PRODUCT SPECIFICATIONS**

NAME

Demi Fraser Broth

### **PRESENTATION**

Dehydrated medium

### STORAGE

10-30°C

### **PACKAGING**

Ref.	Content	Packaging
610091	500 g	500 g of powder in plastic bottle
620091	100 g	100 g of powder in plastic bottle
6100915	5 Kg	5 Kg of powder in plastic bottle

### pH OF THE MEDIUM

7.2 ± 0.2

### USE

Demi Fraser Broth is a liquid medium used with supplements for the detection and enumeration of *L. monocytogenes* and *Listeria* spp in food and environmental samples, according to ISO 11290-1 and ISO 11290-2

#### **TECHNIQUE**

Refer to technical sheet of the product

### APPEARANCE OF THE MEDIUM

Powder medium

Appearance: free-flowing, homogeneous

Colour: beige

Ready-to-use medium Appearance: clear Colour: amber

### SHELFLIFE

4 years

# QUALITY CONTROL

1. Control of general characteristics, label and print

2. Microbiological control

Supplement: Half Fraser Supplement Incubation Conditions:  $30 \pm 1^{\circ}C / 25 \pm 1 \text{ h}$ 

Inoculum for productivity: ≤100 CFU

Microorganism		Specification
Listeria monocytogenes serovar 4b + Escherichia coli + Enterococcus faecalis	WDCM 00021 WDCM 00013 WDCM 00009	Blackening of the medium, >10 colonies on O.A. Listeria Agar

Inoculum for selectivity: >103 CFU

Microorganism		Specification
Escherichia coli	WDCM 00013	Total inhibition on TSA
Enterococcus faecalis	WDCM 00009	<100 colonies on TSA

TSA: Tryptic Soy Agar



